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FILE 'HOME' ENTERED AT 10:13:31 ON 24 SEP 2004

=> file .biotech caplus
COST IN U.S. DOLLARS SINCE FILE ENTRY TOTAL SESSION

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FILES 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
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7 FILES IN THE FILE LIST

=> s hydrogel or polyacrylamide or plyurethane
2 FILES SEARCHED...
L1 451324 HYDROGEL OR POLYACRYLAMIDE OR PLYURETHANE

=> s l1 and (probe or oligonucleotide)
L2 12396 L1 AND (PROBE OR OLIGONUCLEOTIDE)

=> s l2 and photocycloaddition
L3 5 L2 AND PHOTOCYCLOADDITION

=> dup rem l3
PROCESSING COMPLETED FOR L3
L4 4 DUP REM L3 (1 DUPLICATE REMOVED)

=> d ibib abs l4 1-4

L4 ANSWER 1 OF 4 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2003:511934 CAPLUS
DOCUMENT NUMBER: 139:65764
TITLE: Use and evaluation of a [2+2] **photocycloaddition** in immobilization of oligonucleotides on a three-dimensional **hydrogel** matrix
INVENTOR(S): Elghanian, Robert; Brush, Charles K.; Xu, Yanzheng
PATENT ASSIGNEE(S): Amersham Biosciences AB, USA
SOURCE: U.S. Pat. Appl. Publ., 11 pp., Cont.-in-part of U.S. Ser. No. 344,620.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 5
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2003124525	A1	20030703	US 2001-928250	20010809
US 6664061	B2	20031216		
US 6372813	B1	20020416	US 1999-344620	19990625
US 2002146730	A1	20021010	US 2001-25185	20011219
US 2003096265	A1	20030522	US 2002-185279	20020628
WO 2003014392	A2	20030220	WO 2002-IB4038	20020809
WO 2003014392	A3	20031106		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.:
US 1999-344620 A2 19990625
US 2000-224070P P 20000809
US 2000-232305P P 20000912
US 2001-928250 A2 20010809

AB The present invention provides solid supports (e.g., glass) and polymer hydrogels (particularly polymer **hydrogel** arrays present on a solid support) comprising one or more reactive sites for the attachment of biomols., as well as biomols. comprising one or more reactive sites for attachment to solid supports and polymer hydrogels. The invention further provides novel compns. and methods for the preparation of biomols., solid supports, and polymer hydrogels comprising reactive sites. The invention also provides for preparation of crosslinked solid supports, polymer hydrogels, and **hydrogel** arrays, wherein one or more biomols. is attached by means of the reactive sites in a **photocycloaddn**. reaction. Advantageously, according to the invention, crosslinking of the **hydrogel** and attachment of biomols. can be done in a single step. Genes having different expression levels were measured simultaneously using biotin-labeled cRNA generated from human placenta, brain, and heart mRNA. The microarray could detect gene expression at 3 copy per cell.

L4 ANSWER 2 OF 4 BIOTECHDS COPYRIGHT 2004 THE THOMSON CORP. on STN
DUPLICATE 1
ACCESSION NUMBER: 2002-14847 BIOTECHDS
TITLE: Performing expression/single nucleotide polymorphism microarray to determine presence of target, by contacting target with microarray formed by attaching **probe** to support by **photocycloaddition** and scanning microarray;
DNA microaray, DNA chip and SNP for human and yeast gene expression analysis
AUTHOR: ELGHANIAN R; BRUSH C K; XU Y
PATENT ASSIGNEE: MOTOROLA INC
PATENT INFO: WO 2002012566 14 Feb 2002
APPLICATION INFO: WO 2000-US24894 9 Aug 2000
PRIORITY INFO: US 2000-232305 12 Sep 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-434869 [46]
AN 2002-14847 BIOTECHDS
AB DERWENT ABSTRACT:
NOVELTY - Performing (M) expression/single nucleotide polymorphism microarray to detect presence of target (I), comprises attaching **probe** which will recognize (I) to polymer-coated support by (2+2) **photocycloaddition** to form microarray (II), contacting aqueous solution of (I) with (II), to form complex between complementing targets and probes, and scanning (II) to determine presence of (I).

DETAILED DESCRIPTION - Performing (M) expression/single nucleotide polymorphism microarray to detect presence of target (I), comprises attaching **probe** which will recognize (I) to polymer-coated support by (2+2) **photocycloaddition** to form microarray (II), contacting aqueous solution of (I) with (II), to form complex between complementing targets and probes, and scanning (II) to determine presence of (I). In (M), the target solution comprises an aqueous buffer solution and the target, and optionally an active enzyme, and a labeled carrier.

BIOTECHNOLOGY - Preferred Method: (M) further comprises application of a **probe** standard to the polymer-coated support. The **probe** and **probe** standard are applied to the polymer-coated support in about equal amounts, on a weight basis. The aqueous target solution further comprises a target standard. The concentration of the target is determined through comparison of the fluorescence intensities of the target and target standard. The target standard is selected from yeast mRNA and bacterial mRNA, or their combination. Scanning occurs in a spectrometer capable of measuring and recording fluorescence intensity and position. The aqueous target solution comprises a buffer capable of maintaining pH from about 6-9, an active enzyme (such as thermosequanase) that is capable of transferring a label to a **probe**/target complex by single base extension, and a fluorescently labeled carrier (such as di-deoxynucleotide triphosphate)

which provides a transferable label to an active enzyme for transfer to a **probe**/target complex by single base extension. The target is a labeled nucleic acid, and the label is selected from Cy-3, Cy-5, Cy-5.5, and ALEXA FLUOR (RTM), preferably Cy-3. (M) further comprises developing the microarray after application of the target solution, where the developing lasts from 1 minute-42 hours, preferably 16 hours. Developing occurs between 30 and 45degreesC, preferably 37degreesC. Developing lasts for 30-60, preferably 40-50 heating/cooling cycles. (M) further comprises washing with an aqueous wash after developing, where the aqueous wash contains a buffer which comprises phosphate and sodium chloride and is capable of maintaining pH from 6-9. The aqueous wash is performed between 40 and 70degreesC, preferably 50 and 60degreesC. The solid support is a material selected from nylon, polystyrene, glass, latex, polypropylene, and activated cellulose, or their, preferably a glass. The polymer coated support is a **hydrogel** microarray formed by crosslinking a **hydrogel** simultaneous with the attachment of **probe**. The **hydrogel** microarray is prepared by first crosslinking a **hydrogel** prior to attachment of the **probe**. A photosensitizer such as anthroquinone-2-sulfonic acid is added during attachment of the **probe**. The polymer is a polymer, reactive polymer, or copolymer made of at least two co-monomers where at least one of the co-monomers can undergo (2+2) **photocycloaddition**, or a copolymer chemically modified to contain a reactive group that undergoes (2+2) **photocycloaddition**. The polymer or reactive prepolymer contains **polyacrylamide**. The **probe** comprises a nucleic acid fragment containing less than about 1000 nucleotides, and further optionally comprises a linker which is an organic chain of about 6-100 atoms in length. The nucleic acid fragment is selected from synthetic nucleotides and modified nucleotides or their combinations. The **probe** is cDNA, and is chemically modified to contain a reactive group that undergoes (2+2) **photocycloaddition**. The **probe** is chemically modified with a phosphoramidite which is chemically functionalized with a reactive site capable of undergoing (2+2) **photocycloaddition**. The phosphoramidite is functionalized with a cinnamide. The **probe** inherently contains a reactive site that undergoes (2+2) **photocycloaddition**. The reactive site present on the polymer and/or the reactive site present on the **probe**(s) contains an alkene group. The reactive site present on the polymer and/or the reactive site present on the **probe** is selected from dimethyl maleimide, maleimide, thymine, polythymine, acrylate, cinnamate, and citraconimide or their combinations.

USE - (M) is useful for performing expression microarray or single nucleotide polymorphism microarray to determine the presence of a target, where the target is a labeled nucleic acid selected from mRNA, RNA, DNA, amplified RNA, amplified DNA, or its modifications, preferably mRNA, RNA, or DNA (claimed). (M) is useful for performing gene analyses including expression and single nucleotide polymorphism.

ADVANTAGE - (M) is a sensitive method for performing expression microarray or single nucleotide polymorphism microarray to determine the presence of a target.

EXAMPLE - The cRNA targets for gene expression monitoring on expression microarray chip were either total RNA or poly(A) mRNA that were amplified and biotin-labeled. poly(A) RNA were converted into double-strand cDNA using T7-d (T)24 oligo primer and SUPERSCRIPT (RTM) choice system. In vitro transcription was performed on those T7 promoter added dsDNA by using T7 transcriptase. The biotin labeled cRNA was purified and quantitated. The expression chips were then hybridized using biotin labeled cRNA targets in the concentration of 0.08 microg/microl of buffer containing MOTOROLA HYBRIDIZATION (RTM) buffer. The array was then washed with an aqueous buffer containing TRIZMA (RTM), sodium chloride and TWEEN-20 (RTM). The chips were then scanned with a Axon Series A scanner. The gene expression assay was performed using biotin-labeled cRNA generated from human placenta, brain, and heart mRNA. Ten 30 mer human gene expression probes which gave different expression levels and

ten yeast probes were built on the chip. The targets were prepared using human mRNA with different radios of yeast mRNA added for monitoring the sensitivity and dynamic range of the platform performances. The microarray detected gene expression at three copy per cell sensitivity.(34 pages)

L4 ANSWER 3 OF 4 BIOTECHDS COPYRIGHT 2004 THE THOMSON CORP. on STN
ACCESSION NUMBER: 2003-10657 BIOTECHDS
TITLE: Detection of target nucleic acid or protein on chip based DNA microarrays, involves contacting first member of binding pair with second member of binding pair comprising fluorophore, and detecting the fluorophore;
DNA microarray for target nucleic acid or protein detection
AUTHOR: LIU C; MAZUMDER A; BRUSH C K; JOHNSON W T
PATENT ASSIGNEE: LIU C; MAZUMDER A; BRUSH C K; JOHNSON W T
PATENT INFO: US 2002146730 10 Oct 2002
APPLICATION INFO: US 2001-25185 19 Dec 2001
PRIORITY INFO: US 2001-25185 19 Dec 2001; US 1999-344620 25 Jun 1999
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2003-237971 [23]
AN 2003-10657 BIOTECHDS
AB DERWENT ABSTRACT:
NOVELTY - Detecting a target nucleic acid or protein, comprising providing a target containing a nucleic acid or protein and a first member of a binding pair; hybridizing the target to a **probe** attached to a **hydrogel** matrix through a 2+2 **photocycloaddition**, contacting the first member of the binding pair with a second member of the binding pair comprising a fluorophore, and detecting the fluorophore, is new.

DETAILED DESCRIPTION - An INDEPENDENT CLAIM is included for a kit for detecting a target nucleic acid or protein comprising a **hydrogel** matrix, a **probe** and two members of binding pair.

BIOTECHNOLOGY - Preferred Method: The nucleic acid is synthesized by producing cDNA from mRNA, cDNA or cRNA from a DNA. The target is synthesized by incorporating the first member of a binding pair into the nucleic acid by polymerization. At least one of the second members of the binding pair is contacted with an antibody comprising a first member. Preferred Component: The first member comprises biotin (preferred), digoxygenin, or bromouridine. The second member comprises avidin, streptavidin (preferred), biotin antibody, digoxygenin antibody, or bromouridine antibody. The protein is cell lysate. The **probe** comprises a reactive site capable of undergoing a 2+2 **photocycloaddition**. The **hydrogel** matrix comprises a reactive site capable of undergoing a 2+2 **photocycloaddition**. The streptavidin is attached preferably to three to four fluorophore. The fluorophore is cyanine dyes or ALEXA FLOUR (RTM) dyes. The cyanine dye is Cy-3, Cy-5, or Cy-5.5. The ALEXA FLOUR (RTM) dye is ALEXA-532 (RTM), ALEXA-647 (RTM) (preferred), or ALEXA-633. The antibody is biotinylated anti-streptavidin antibody.

USE - For the detection of target nucleic acid or protein on chip based DNA microarrays.

ADVANTAGE - The invention provides a high-sensitivity target detection methods for use with **hydrogel** microarrays that provide a good signal to noise ratio. (12 pages)

L4 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2001:12732 CAPLUS
DOCUMENT NUMBER: 134:68455
TITLE: Methods and compositions for attachment of biomolecules to solid supports, hydrogels, and **hydrogel** arrays

INVENTOR(S): Johnson, Travis; McGowen, John; Beuhler, Allyson; Brush, Charles Kimball; Lajos, Robert Emil
PATENT ASSIGNEE(S): Motorola Inc., USA
SOURCE: PCT Int. Appl., 46 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 5
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001001143	A2	20010104	WO 2000-US17422	20000623
WO 2001001143	A3	20010308		
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
US 6372813	B1	20020416	US 1999-344620	19990625
EP 1190254	A2	20020327	EP 2000-941693	20000623
EP 1190254	B1	20040915		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
JP 2003524150	T2	20030812	JP 2001-507097	20000623
AU 768326	B2	20031211	AU 2000-56362	20000623
US 2003078314	A1	20030424	US 2001-976986	20011011
US 6686161	B2	20040203		

PRIORITY APPLN. INFO.:
US 1999-344620 A 19990625
WO 2000-US17422 W 20000623

AB The present invention provides solid supports (e.g., glass) and polymer hydrogels (particularly polymer **hydrogel** arrays present on a solid support) comprising one or more reactive sites for the attachment of biomols., as well as biomols. comprising one or more reactive sites for attachment to solid supports and polymer hydrogels. The invention further provides novel compns. and methods for the preparation of biomols., solid supports and polymer hydrogels comprising reactive sites. The invention also provides for preparation of crosslinked solid supports, polymer hydrogels, and **hydrogel** arrays, wherein one or more biomols. is attached by means of the reactive sites in a **photocycloaddn**. reaction. Advantageously, according to the invention, crosslinking of the **hydrogel** and attachment of biomols. can be done in a single step. Photopolymer **polyacrylamide** co-N-(6-acryloylhexyl)-2,3-dimethylmaleimide was prepared This polymer is coated on a solid support and exposed to UV radiation to photocrosslink and form a **hydrogel** . Unreacted maleimide functional groups in the **hydrogel** are then reacted with maleimide-functionalized DNA **oligonucleotide**.

=> d his

(FILE 'HOME' ENTERED AT 10:13:31 ON 24 SEP 2004)

FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS' ENTERED AT 10:13:45 ON 24 SEP 2004

L1 451324 S HYDROGEL OR POLYACRYLAMIDE OR PLYURETHANE
L2 12396 S L1 AND (PROBE OR OLIGONUCLEOTIDE)
L3 5 S L2 AND PHOTOCYCLOADDITION
L4 4 DUP REM L3 (1 DUPLICATE REMOVED)

```
=> s l1 and photocycloaddition
L5          11 L1 AND PHOTOCYCLOADDITION

=> dup rem l5
PROCESSING COMPLETED FOR L5
L6             8 DUP REM L5 (3 DUPLICATES REMOVED)

=> d ibib abs l6 1-8

L6   ANSWER 1 OF 8  BIOSIS  COPYRIGHT (c) 2004 The Thomson Corporation.  on STN
ACCESSION NUMBER:   2004:150375  BIOSIS
DOCUMENT NUMBER:    PREV200400153772
TITLE:              Methods and compositions for attachment of biomolecules to
                    solid supports, hydrogels, and hydrogel arrays.
AUTHOR(S):          Johnson, Travis [Inventor, Reprint Author]; McGowen, John
                    [Inventor]; Beuhler, Allyson [Inventor]; Brush, Charles
                    Kimball [Inventor]; Lajos, Robert Emil [Inventor]
CORPORATE SOURCE:   ASSIGNEE: Amersham Biosciences AB, Uppsala, Sweden
PATENT INFORMATION: US 6686161 February 03, 2004
SOURCE:             Official Gazette of the United States Patent and Trademark
                    Office Patents, (Feb 3 2004) Vol. 1279, No. 1.
                    http://www.uspto.gov/web/menu/patdata.html. e-file.
                    ISSN: 0098-1133 (ISSN print).
DOCUMENT TYPE:      Patent
LANGUAGE:           English
ENTRY DATE:         Entered STN: 17 Mar 2004
                    Last Updated on STN: 17 Mar 2004
AB   The present invention provides solid supports (e.g., glass) and polymer
     hydrogels (particularly polymer hydrogel arrays present on a
     solid support) comprising one or more reactive sites for the attachment of
     biomolecules, as well as biomolecules comprising one or more reactive
     sites for attachment to solid supports and polymer hydrogels.  The
     invention further provides novel compositions and methods for the
     preparation of biomolecules, solid supports, and polymer hydrogels
     comprising reactive sites.  The invention also provides for preparation of
     crosslinked solid supports, polymer hydrogels, and hydrogel
     arrays, wherein one or more biomolecules is attached by means of the
     reactive sites in a photocycloaddition reaction.
     Advantageously, according to the invention, crosslinking of the
     hydrogel and attachment of biomolecules can be done in a single
     step.

L6   ANSWER 2 OF 8  CAPLUS  COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER:        2003:511934  CAPLUS
DOCUMENT NUMBER:         139:65764
TITLE:                   Use and evaluation of a [2+2]
                         photocycloaddition in immobilization of
                         oligonucleotides on a three-dimensional
                         hydrogel matrix
INVENTOR(S):             Elghanian, Robert; Brush, Charles K.; Xu, Yanzheng
PATENT ASSIGNEE(S):      Amersham Biosciences AB, USA
SOURCE:                  U.S. Pat. Appl. Publ., 11 pp., Cont.-in-part of U.S.
                         Ser. No. 344,620.
                         CODEN: USXXCO
DOCUMENT TYPE:           Patent
LANGUAGE:                English
FAMILY ACC. NUM. COUNT:  5
PATENT INFORMATION:

     PATENT NO.          KIND   DATE        APPLICATION NO.          DATE
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     US 2003124525        A1    20030703    US 2001-928250           20010809
     US 6664061           B2    20031216
     US 6372813           B1    20020416    US 1999-344620           19990625
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US 2002146730 A1 20021010 US 2001-25185 20011219
US 2003096265 A1 20030522 US 2002-185279 20020628
WO 2003014392 A2 20030220 WO 2002-IB4038 20020809
WO 2003014392 A3 20031106

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 1999-344620 A2 19990625
US 2000-224070P P 20000809
US 2000-232305P P 20000912
US 2001-928250 A2 20010809

AB The present invention provides solid supports (e.g., glass) and polymer hydrogels (particularly polymer **hydrogel** arrays present on a solid support) comprising one or more reactive sites for the attachment of biomols., as well as biomols. comprising one or more reactive sites for attachment to solid supports and polymer hydrogels. The invention further provides novel compns. and methods for the preparation of biomols., solid supports, and polymer hydrogels comprising reactive sites. The invention also provides for preparation of crosslinked solid supports, polymer hydrogels, and **hydrogel** arrays, wherein one or more biomols. is attached by means of the reactive sites in a **photocycloaddn**. reaction. Advantageously, according to the invention, crosslinking of the **hydrogel** and attachment of biomols. can be done in a single step. Genes having different expression levels were measured simultaneously using biotin-labeled cRNA generated from human placenta, brain, and heart mRNA. The microarray could detect gene expression at 3 copy per cell.

L6 ANSWER 3 OF 8 BIOTECHDS COPYRIGHT 2004 THE THOMSON CORP. on STN
DUPLICATE 1
ACCESSION NUMBER: 2002-14847 BIOTECHDS
TITLE: Performing expression/single nucleotide polymorphism microarray to determine presence of target, by contacting target with microarray formed by attaching probe to support by **photocycloaddition** and scanning microarray; DNA microaray, DNA chip and SNP for human and yeast gene expression analysis
AUTHOR: ELGHANIAN R; BRUSH C K; XU Y
PATENT ASSIGNEE: MOTOROLA INC
PATENT INFO: WO 2002012566 14 Feb 2002
APPLICATION INFO: WO 2000-US24894 9 Aug 2000
PRIORITY INFO: US 2000-232305 12 Sep 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-434869 [46]
AN 2002-14847 BIOTECHDS
AB DERWENT ABSTRACT:
NOVELTY - Performing (M) expression/single nucleotide polymorphism microarray to detect presence of target (I), comprises attaching probe which will recognize (I) to polymer-coated support by (2+2) **photocycloaddition** to form microarray (II), contacting aqueous solution of (I) with (II), to form complex between complementing targets and probes, and scanning (II) to determine presence of (I).
DETAILED DESCRIPTION - Performing (M) expression/single nucleotide polymorphism microarray to detect presence of target (I), comprises attaching probe which will recognize (I) to polymer-coated support by (2+2) **photocycloaddition** to form microarray (II), contacting aqueous solution of (I) with (II), to form complex between complementing

targets and probes, and scanning (II) to determine presence of (I). In (M), the target solution comprises an aqueous buffer solution and the target, and optionally an active enzyme, and a labeled carrier.

BIOTECHNOLOGY - Preferred Method: (M) further comprises application of a probe standard to the polymer-coated support. The probe and probe standard are applied to the polymer-coated support in about equal amounts, on a weight basis. The aqueous target solution further comprises a target standard. The concentration of the target is determined through comparison of the fluorescence intensities of the target and target standard. The target standard is selected from yeast mRNA and bacterial mRNA, or their combination. Scanning occurs in a spectrometer capable of measuring and recording fluorescence intensity and position. The aqueous target solution comprises a buffer capable of maintaining pH from about 6-9, an active enzyme (such as thermosequanase) that is capable of transferring a label to a probe/target complex by single base extension, and a fluorescently labeled carrier (such as di-deoxynucleotide triphosphate) which provides a transferable label to an active enzyme for transfer to a probe/target complex by single base extension. The target is a labeled nucleic acid, and the label is selected from Cy-3, Cy-5, Cy-5.5, and ALEXA FLUOR (RTM), preferably Cy-3. (M) further comprises developing the microarray after application of the target solution, where the developing lasts from 1 minute-42 hours, preferably 16 hours. Developing occurs between 30 and 45degreesC, preferably 37degreesC. Developing lasts for 30-60, preferably 40-50 heating/cooling cycles. (M) further comprises washing with an aqueous wash after developing, where the aqueous wash contains a buffer which comprises phosphate and sodium chloride and is capable of maintaining pH from 6-9. The aqueous wash is performed between 40 and 70degreesC, preferably 50 and 60degreesC. The solid support is a material selected from nylon, polystyrene, glass, latex, polypropylene, and activated cellulose, or their, preferably a glass. The polymer coated support is a **hydrogel** microarray formed by crosslinking a **hydrogel** simultaneous with the attachment of probe. The **hydrogel** microarray is prepared by first crosslinking a **hydrogel** prior to attachment of the probe. A photosensitizer such as anthroquinone-2-sulfonic acid is added during attachment of the probe. The polymer is a polymer, reactive polymer, or copolymer made of at least two co-monomers where at least one of the co-monomers can undergo (2+2) **photocycloaddition**, or a copolymer chemically modified to contain a reactive group that undergoes (2+2) **photocycloaddition**. The polymer or reactive prepolymer contains **polyacrylamide**. The probe comprises a nucleic acid fragment containing less than about 1000 nucleotides, and further optionally comprises a linker which is an organic chain of about 6-100 atoms in length. The nucleic acid fragment is selected from synthetic nucleotides and modified nucleotides or their combinations. The probe is cDNA, and is chemically modified to contain a reactive group that undergoes (2+2) **photocycloaddition**. The probe is chemically modified with a phosphoramidite which is chemically functionalized with a reactive site capable of undergoing (2+2) **photocycloaddition**. The phosphoramidite is functionalized with a cinnamide. The probe inherently contains a reactive site that undergoes (2+2) **photocycloaddition**. The reactive site present on the polymer and/or the reactive site present on the probe(s) contains an alkene group. The reactive site present on the polymer and/or the reactive site present on the probe is selected from dimethyl maleimide, maleimide, thymine, polythymine, acrylate, cinnamate, and citraconimide or their combinations.

USE - (M) is useful for performing expression microarray or single nucleotide polymorphism microarray to determine the presence of a target, where the target is a labeled nucleic acid selected from mRNA, RNA, DNA, amplified RNA, amplified DNA, or its modifications, preferably mRNA, RNA, or DNA (claimed). (M) is useful for performing gene analyses including expression and single nucleotide polymorphism.

ADVANTAGE - (M) is a sensitive method for performing expression

microarray or single nucleotide polymorphism microarray to determine the presence of a target.

EXAMPLE - The cRNA targets for gene expression monitoring on expression microarray chip were either total RNA or poly(A) mRNA that were amplified and biotin-labeled. poly(A) RNA were converted into double-strand cDNA using T7-d (T)24 oligo primer and SUPERSCRIPT (RTM) choice system. In vitro transcription was performed on those T7 promoter added dsDNA by using T7 transcriptase. The biotin labeled cRNA was purified and quantitated. The expression chips were then hybridized using biotin labeled cRNA targets in the concentration of 0.08 microg/microl of buffer containing MOTOROLA HYBRIDIZATION (RTM) buffer. The array was then washed with an aqueous buffer containing TRIZMA (RTM), sodium chloride and TWEEN-20 (RTM). The chips were then scanned with a Axon Series A scanner. The gene expression assay was performed using biotin-labeled cRNA generated from human placenta, brain, and heart mRNA. Ten 30 mer human gene expression probes which gave different expression levels and ten yeast probes were built on the chip. The targets were prepared using human mRNA with different radios of yeast mRNA added for monitoring the sensitivity and dynamic range of the platform performances. The microarray detected gene expression at three copy per cell sensitivity.(34 pages)

L6 ANSWER 4 OF 8 BIOTECHDS COPYRIGHT 2004 THE THOMSON CORP. on STN
DUPLICATE 2
ACCESSION NUMBER: 2003-10657 BIOTECHDS
TITLE: Detection of target nucleic acid or protein on chip based DNA microarrays, involves contacting first member of binding pair with second member of binding pair comprising fluorophore, and detecting the fluorophore;
DNA microarray for target nucleic acid or protein detection
AUTHOR: LIU C; MAZUMDER A; BRUSH C K; JOHNSON W T
PATENT ASSIGNEE: LIU C; MAZUMDER A; BRUSH C K; JOHNSON W T
PATENT INFO: US 2002146730 10 Oct 2002
APPLICATION INFO: US 2001-25185 19 Dec 2001
PRIORITY INFO: US 2001-25185 19 Dec 2001; US 1999-344620 25 Jun 1999
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2003-237971 [23]
AN 2003-10657 BIOTECHDS
AB DERWENT ABSTRACT:
NOVELTY - Detecting a target nucleic acid or protein, comprising providing a target containing a nucleic acid or protein and a first member of a binding pair; hybridizing the target to a probe attached to a **hydrogel** matrix through a 2+2 **photocycloaddition**, contacting the first member of the binding pair with a second member of the binding pair comprising a fluorophore, and detecting the fluorophore, is new.

DETAILED DESCRIPTION - An INDEPENDENT CLAIM is included for a kit for detecting a target nucleic acid or protein comprising a **hydrogel** matrix, a probe and two members of binding pair.

BIOTECHNOLOGY - Preferred Method: The nucleic acid is synthesized by producing cDNA from mRNA, cDNA or cRNA from a DNA. The target is synthesized by incorporating the first member of a binding pair into the nucleic acid by polymerization. At least one of the second members of the binding pair is contacted with an antibody comprising a first member. Preferred Component: The first member comprises biotin (preferred), digoxygenin, or bromouridine. The second member comprises avidin, streptavidin (preferred), biotin antibody, digoxygenin antibody, or bromouridine antibody. The protein is cell lysate. The probe comprises a reactive site capable of undergoing a 2+2 **photocycloaddition**. The **hydrogel** matrix comprises a reactive site capable of undergoing a 2+2 **photocycloaddition**. The streptavidin is attached preferably to three to four fluorophore. The fluorophore is

cyanine dyes or ALEXA FLOUR (RTM) dyes. The cyanine dye is Cy-3, Cy-5, or Cy-5.5. The ALEXA FLOUR (RTM) dye is ALEXA-532 (RTM), ALEXA-647 (RTM) (preferred), or ALEXA-633. The antibody is biotinylated anti-streptavidin antibody.

USE - For the detection of target nucleic acid or protein on chip based DNA microarrays.

ADVANTAGE - The invention provides a high-sensitivity target detection methods for use with **hydrogel** microarrays that provide a good signal to noise ratio. (12 pages)

L6 ANSWER 5 OF 8 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2002:286636 BIOSIS
DOCUMENT NUMBER: PREV200200286636
TITLE: Methods and compositions for attachment of biomolecules to solid supports, hydrogels, and **hydrogel** arrays.
AUTHOR(S): Johnson, Travis [Inventor, Reprint author]; McGowen, John [Inventor]; Beuhler, Allyson [Inventor]; Brush, Charles Kimball [Inventor]; Lajos, Robert Emil [Inventor]
CORPORATE SOURCE: Chandler, AZ, USA
ASSIGNEE: Motorola, Schaumburg, IL, USA
PATENT INFORMATION: US 6372813 April 16, 2002
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Apr. 16, 2002) Vol. 1257, No. 3. http://www.uspto.gov/web/menu/patdata.html. e-file. CODEN: OGUPE7. ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
ENTRY DATE: Entered STN: 8 May 2002
Last Updated on STN: 8 May 2002

AB The present invention provides solid supports (e.g., glass) and polymer hydrogels (particularly polymer **hydrogel** arrays present on a solid support) comprising one or more reactive sites for the attachment of biomolecules, as well as biomolecules comprising one or more reactive sites for attachment to solid supports and polymer hydrogels. The invention further provides novel compositions and methods for the preparation of biomolecules, solid supports, and polymer hydrogels comprising reactive sites. The invention also provides for preparation of crosslinked solid supports, polymer hydrogels, and **hydrogel** arrays, wherein one or more biomolecules is attached by means of the reactive sites in a **photocycloaddition** reaction. Advantageously, according to the invention, crosslinking of the **hydrogel** and attachment of biomolecules can be done in a single step.

L6 ANSWER 6 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:638302 CAPLUS
DOCUMENT NUMBER: 137:170343
TITLE: Hydrogels and **hydrogel** arrays made from **polyacrylamide**-based reactive prepolymers crosslinked by [2+2] cycloaddition
INVENTOR(S): Beuhler, Allyson; McGowen, John
PATENT ASSIGNEE(S): Motorola, Inc., USA
SOURCE: U.S. Pat. Appl. Publ., 14 pp., Cont.-in-part of U.S. 6,391,937.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2002115740	A1	20020822	US 2002-131426	20020423
US 2002035167	A1	20020321	US 1999-344217	19990625

US 6391937 B2 20020521
PRIORITY APPLN. INFO.: US 1998-109821P P 19981125
US 1999-344217 A2 19990625

AB The title prepolymers (A) comprise a copolymer of a first monomer such as acrylamide, a [2+2] photocyclizable monomer, e.g., N-(6-acryloyloxyhexyl)-2,3-dimethylmaleimide, vinyl cinnamate, etc., and a second monomer selected from acrylic acid, glycidyl methacrylate, methacrylic acid, or mixture thereof, and undergo [2+2] cycloaddn. to be crosslinked upon exposure to UV light to form a **hydrogel**. Thus, polymerizing 17.06 g acrylamide and 3.35 g N-(6-acryloyloxyhexyl)-2,3-dimethylmaleimide in the presence of 0.39 g copper(II) sulfate pentahydrate and 0.3 g potassium peroxodisulfate gave an A, a 20% solid content of which was coated on a solid support with 1% photosensitizer (anthroquinone 2-sulfonic acid sodium salt) and exposed with UV radiation to give an array pattern of crosslinked porous **hydrogel** after removing developer solution

L6 ANSWER 7 OF 8 BIOTECHDS COPYRIGHT 2004 THE THOMSON CORP. on STN
DUPLICATE 3
ACCESSION NUMBER: 2001-06902 BIOTECHDS
TITLE: Composition for attachment of biomolecules to solid supports, polymer hydrogels, and **hydrogel** arrays, comprises using **photocycloaddition** between reactive sites on the support and biomolecules;
DNA immobilization to solid support
AUTHOR: Johnson T; McGowen J; Beuhler A; Brush C K; Lajos R E
PATENT ASSIGNEE: Motorola
LOCATION: Schaumburg, IL, USA.
PATENT INFO: WO 2001001143 4 Jan 2001
APPLICATION INFO: WO 2000-US17422 23 Jun 2000
PRIORITY INFO: US 1999-344620 25 Jun 1999
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2001-168320 [17]
AN 2001-06902 BIOTECHDS
AB A composition comprising solid supports, optionally polymer-coated, attached to biomolecules by 2 + 2 cycloaddition between reactive sites on the support or polymer and the biomolecules is new. The biomolecule comprises a nucleic acid fragment containing less than 1,000 nucleotides and optionally further contains a spacer region. The solid support is nylon, polystyrene, glass, latex, polypropylene or activated cellulose, e.g. a bead, membrane, microwell, centrifuge tube or slide. In an example, copolymer **polyacrylamide** coglycidyl methacrylate was modified with acrylic acid to form a photoreactive **polyacrylamide** reactive prepolymer. This was coated on a solid support and exposed to UV radiation to photocrosslink in an array pattern of 100 um diameter pads spaced at 300 um pitch. The unexposed polymer was washed away, leaving a grid of **hydrogel** pads. The pads contained unreacted acrylate functional groups as attachment sites for biomolecules. (46pp)

L6 ANSWER 8 OF 8 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 1989:47316 BIOSIS
DOCUMENT NUMBER: PREV198987023316; BA87:23316
TITLE: ON THE DNA BENDING BY PSORALEN INTERSTRAND CROSSLINKING A GEL ELECTROPHORETIC STUDY.
AUTHOR(S): ZHEN W-P [Reprint author]; DAHL O; BUCHARDT O; NIELSEN P E
CORPORATE SOURCE: DEP BIOCHEMISTRY B, PANUM INST, UNIV COPENHAGEN, BLEDGAMSVEJ 3, DK-220 COPENHAGEN N DENMARK
SOURCE: Photochemistry and Photobiology, (1988) Vol. 48, No. 5, pp. 643-646.
CODEN: PHCBAP. ISSN: 0031-8655.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 7 Jan 1989

Last Updated on STN: 7 Jan 1989

AB A synthetic, partially double stranded decadeoxyribonucleotide with cohesive ends, containing one potential psoralen photo-crosslinking site centrally positioned (5'-d(CGGGCTACCC)), + 3'- d(CCGATGGGGC). has been ligated to double stranded DNA oligomers, which were subsequently photoreacted with 4,5'-8-timethylpsoralen. It was found that psoralen DNA interstranded crosslinking does not significantly alter the electrophoretic mobility of these DNA molecules in **polyacrylamide** gels. Based on this, we conclude that any bends in the DNA helix that may be induced by psoralen DNA interstrand crosslinking must be significantly less than the 45° proposed by Tomic et al. (1987) (Science, 238, 1722) and/or a different nature than the DNA sequence dependent bends due to d(A)n tracts.

=> FIL STNGUIDE

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CA SUBSCRIBER PRICE	-2.80	-2.80

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LAST RELOADED: Sep 17, 2004 (20040917/UP).